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ON THE SPECIFIC GRAVITY OF SPIROSTOMUM, PARAMÆCIUM, AND THE TADPOLE IN RELATION TO THE PROBLEM OF GEOTAXIS.[1]

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It is known that some algæ and Infusoria tend to collect near the surface of the water in which they live. A number of experiments have been made which seem to demonstrate that the tendency thus manifest cannot be ascribed altogether to the attraction of light, or to the source of oxygen supply, since these algæ and Infusoria still move upwards when the experimental conditions are such as destroy or reverse the normal relations to air and light. It is, therefore, affirmed that these organisms react to the force of gravity. This reaction, or negative geotaxis, is attributed by Schwarz ('84) to the direct influence of gravity on the organism, which incites motion in the opposed direction, while Jensen ('93) attributes the reaction to the indirect influence of gravity on the organism by means of the difference in hydrostatic pressure at different depths. The experiments of these authors seem to show that the possibly excentric position of the center of gravity in the organism cannot be regarded as a factor determining the direction of motion.

Dr. C. B. Davenport suggested that a simple and hitherto unrecorded method of approaching the problem of geotaxis might be by means of solutions of varying density in which the infusorian would still live. Obviously, if the conclusion of Schwarz ('84) were right, the negatively geotactic organism should become positively geotactic in solutions of greater specific gravity than its own, supposing the animal to be normally heavier than water.

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[1] This short study was undertaken as laboratory work in connection with Dr. C. B. Davenport's course in *Experimental Morphology*. My cordial thanks are due Dr. Davenport for the kind interest with which he followed the work.

Unfortunately, for the success of the experiment, I have been unable to obtain in Cambridge, Paramæcia which showed decided geotactic reaction. My results may, nevertheless, serve as a basis for further experiment by those more fortunate in the material at their command.

The Method.

The specific gravity of the organism was to be obtained by finding the density of a solution in which the animal, either dead or paralyzed, remained suspended without rising or falling. This known, the movements of the living infusorian in solutions of the same, of greater, and of less specific gravity were to be recorded. It was, therefore, necessary to find a substance in solutions of which Infusoria will live, and a reagent that will kill or paralyze the animal with little apparent change of form.

I found that in solutions of salt, sugar, and glucose, of sufficient density to be serviceable, Infusoria do not live, but in dying increase their specific gravity, probably owing in great part to loss of water. Gelatine was not satisfactory, since even weak solutions tend to become stiff when cold. After failing with the above-mentioned substances, I found gum arabic admirably adapted to my purpose. Solutions surpassing the specific gravity of protoplasm are easily obtained, and Paramæcia or Spirostoma live for hours in solutions heavier than their specific gravity, not only surviving, but also multiplying, when kept for days or weeks in solutions of less weight.

To exclude the small error that might arise from water contained in the gum, the specific gravity of the solution has not been determined by adding a known weight of the gum to a given volume of water, but in each case the solution has been weighed and compared directly with the weight of the same volume of water.

Spirostomum.

Although Spirostoma frequently lie on the bottom of the vessel in which they are cultivated, they are also often found suspended in the water with their long axis in a vertical plane, and with the anterior end of the body directed upwards. As it is possible that the maintenance of this position is in reaction to gravity, I tried the effect of placing Spirostoma in solutions denser than water. When Spirostomum moves, it usually advances first the smaller end of its elongated body, or the end which is opposite the large contractile vacuole; and this I consequently call the anterior end of the body. Spirostomum occasionally reverses its cilia and moves backwards, but the regression is of momentary duration, and the small organism returns directly to its usual manner of motion.

A fortunate accident by which a faint trace of some unknown poison must have been present in the culture jar to which a number of Spirostoma were transferred, rendered these Infusoria motionless without killing them. When examined under the microscope it was found that their cilia did not move, yet an occasional spasmodic contraction demonstrated that life still continued. I was thus enabled to obtain the weight of *living* Infusoria, and the result was subsequently confirmed by similar experiments with Spirostoma that were not poisoned, but merely exhausted by long cultivation in the laboratory.

To avoid currents that would arise from introducing water with the Spirostoma into the gum arabic solution, they were first drawn from the culture jar with a pipette and ejected into a watch glass, which was gently shaken so that the Spirostoma gathered at the bottom. The water that had been taken up with them was then withdrawn, and gum arabic of the same density as the solution to which they were about to be transferred was substituted. In this solution of gum arabic the Spirostoma were again drawn into a pipette and ejected gently into the midst of the solution with which their specific gravity was to be compared.

In determining the specific gravity of these Infusoria, the reaction during the first fifteen or twenty minutes is alone to be trusted, since Spirostoma, and also Paramæcia, whether living or dead, increase their specific gravity on remaining long in a solution of greater density than water. The paralyzed, but still living Spirostoma were placed in solutions of differing densities with the following result :

In H_2O, every Spirostomum immediately fell to the bottom.

In gum arabic solutions of sp. grav. 1.0110, all of the Spirostoma fell to the bottom within fifteen minutes, except two or three that were still falling.

sp. grav. 1.0165, the Spirostoma were carried by the slight currents. For the first five minutes indifferently. They then slowly rose, but not to the surface, then fell, but not to the bottom. They were evidently nearly balanced.

" " 1.0165, the above experiment was repeated, and the Spirostoma remained scattered for twenty to thirty minutes. Then all drifted upwards except six which lay on the bottom.

" " 1.0180, Spirostoma remain still for a long time.

" " 1.0190, Spirostoma remain still for a short time, and then rise, *several coming to the surface.*

" " 1.0190, the above experiment was repeated with the same result.

" " 1.0208, Spirostoma slowly rise.

" " 1.0230, after ten minutes thirteen Spirostoma were floating on the surface, and but two still remained in the solution. Both of these were rising. When the solution was shaken, all were scattered, and then rose to the surface again. After two hours all fell to the bottom.

From these experiments it appears that the specific gravity of Spirostomum lies between 1.0165 and 1.0180, or very near to 1.017.

Jensen ('93) also attempted to obtain the specific gravity of an infusorian, Paramæcium, by finding the density of a solution in which the infusorian would neither rise nor fall. He chose for the experiment, however, potassium carbonate, which immediately kills the organism. By means of solutions of potassium carbonate, Jensen decided that the specific gravity of Paramæcium was 1.25. Since his conclusions differ so greatly from the results obtained with gum arabic and living Spirostoma, I repeated the experiment by placing Spirostoma in a solution of potassium carbonate of 1.25 specific gravity, and found that this infusorian rose at once to the surface, and then fell directly back into the solution, where it remained in fact suspended. Evidently potassium carbonate of this density rapidly reduces the protoplasm of the organism to its own weight. Only the extent of the change (from 1.017 to 1.25) was surprising, for I had already found that Spirostomum changed its weight slightly when living in a medium denser than water, and had also found that, when killed with any one of several reagents, the specific gravity of Spirostomum increases. For example, when Spirostomum is killed with quinine, it sinks in solutions of gum arabic of 1.028 specific gravity, although the living but motionless Spirostomum quickly rises to the surface in a solution of this density.

On placing Spirostomum in a solution of gum arabic slightly denser than its own protoplasm, I found that the infusorian did not remain still in a vertical position, as is its habit in water, but wandered restlessly about. Of those individuals which at the time of observation occupied a vertical, or nearly vertical position, the number with the anterior extremity directed upwards was 48. The number of those with the anterior extremity directed downwards was 81. The larger number of individuals having their anterior extremity directed downwards suggests a tendency to reverse the normal position maintained in water, where the anterior extremity is directed upwards. I doubt, however, if the difference in the numbers should be thus interpreted; for if Spirostomum merely wandered back and forth between the upper and lower surfaces of the liquid, it would rise in a dense solution more rapidly than it would descend. There would thus be at any moment more individuals going downwards than upwards. The power of nice adjustment between the force of the cilia and the force of gravity, which maintains Spirostomum suspended in water, was lost in those individuals which I observed when placed in solutions of greater specific gravity than their own. My conclusions regarding their reaction to gravity are, therefore, purely negative.

Paramœcium.

To obtain the specific gravity of Paramæcia, I tried to paralyze them with ether, quinine, and nicotine, but invariably killed the Paramæcia, which in dying went to pieces. I therefore added a few drops of 0.5 per cent acetic acid to the water containing the Infusoria. As acetic acid is said to swell protoplasm, I also killed Paramæcia by the fumes of osmic acid. Both these reagents caused the Paramæcia to change their shape somewhat in dying, by becoming wider and shorter. Their form, however, remained symmetrical in both cases, and seemed neither enlarged nor distorted.

The results obtained by the two methods were similar. When the killed Paramæcia were placed in a gum arabic solution of 1.018 specific gravity, they remained long suspended in the solution. In a solution of 1.024 specific gravity they rose to the surface as rapidly as they fell when placed in water. The reaction in solutions of nearly the same specific gravity as Paramæcium is less certain than with Spirostomum, since the smaller size of Paramæcium makes it more liable to drift up or down with chance currents, and less rapid in its response to gravity. There is, however, no doubt that the specific gravity of Paramæcium thus killed differs but little from that of living Spirostomum, and differs very greatly from the specific gravity assigned this infusorian by Jensen ('93).

The Paramæcia found in the cultures of our laboratory and in the water of the pools about Cambridge did not show geotactic tendencies. If one were placed in clear water, and were drawn into a capillary tube held vertical, it would rise to the upper surface of the water drawn into the tube, and if the tube were now directly reversed, the Paramæcium would rise again; but if the tube were retained in its original vertical position, the Paramæcium wandered back and forth between the upper and lower surfaces of the water. The water of the culture vessels, which contained dead leaves, bacteria, etc., soon swarmed throughout with Paramæcia, which were found in greatest quantities at the surface where a scum of bacteria gathered. If these superficial bacteria were placed in a glass of clear water, they wandered up and down indifferently.

Tadpoles.

Some toad tadpoles that had just left their gelatinous envelope were observed by Dr. Davenport to swim at once to the surface of the water in which they were kept. He therefore suggested that the negative geotaxis they thus manifested should also be tested in solutions of gum arabic of differing densities.

I found that between the lengths of 9½ mm. and 12 mm., the specific gravity of the tadpoles decreased from 1.044 for the smaller, to 1.017 for the larger. The smaller tadpoles, which showed marked geotactic tendency, were placed in a phial filled to the brim with a solution of gum arabic heavier than the tadpoles, and the phial was then inverted with care that no bubbles of air were admitted. The tadpoles, which could not have been attracted upwards by the light, since light was admitted at all sides through the glass of the phial, immediately swam upwards, as they had done in water. As there was no air between the bottom of the inverted bottle and the upper surface of the solution, it was equally improbable that the tadpoles were attracted upwards by the air.

When placed in solutions of the same specific gravity as themselves, it was found that the tadpoles still swam to the upper surface of the solution, which as before was in immediate contact with the bottom of the inverted phial. From these experiments it appears that the attraction upwards is neither air nor light, and that the direct action of gravity as expressed in the weight of this organism does not act as the incentive to negative geotaxis. It is possible that gravity may act, however, on some internal organ which is unaffected by the change in the density of the surrounding medium, and may by this means induce the movement upwards. Loeb (**'91**) has shown that the semicircular canals of the ear in sharks act as balancing organs. Should this also be true for tadpoles, we might find here the explanation of their constant motion upwards, even when the direction of the action of gravity on the organism as a whole has been changed by placing the organism, normally heavier than water, in a medium of greater specific gravity than itself.

It is, moreover, by no means impossible that the action of gravity, which determines the direction in which the Infusoria move, may be through the internal organization of the animal.

In this case the density of the surrounding medium might be expected to effect little, if any, change in the direction in which the organism moves.

Results.

(1) The specific gravity of living Spirostomum lies between 1.016 and 1.018, or is near to 1.017.

(2) The specific gravity of Paramæcium killed by acetic acid, or by the fumes of osmic acid, differs little, if any, from the specific gravity of living Spirostomum, and is about 1.017.

(3) Small tadpoles that are negatively geotactic do not become positively geotactic when placed in solutions heavier than their own specific gravity, as one would expect were their upward motion in direct response to the action of gravity on the organism as a whole. These tadpoles show constant negative geotaxis in water, in a solution of their own specific gravity, and in heavier solutions.

RADCLIFFE COLLEGE, CAMBRIDGE,
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LIST OF PAPERS REFERRED TO.

JENSEN, P. **'93**. Die absolute Kraft einer Flimmerzelle. *Arch. f. d. ges. Physiol.* Vol. liv, pp. 537–551. June 24, 1893.

LOEB, J. **'91**. Ueber Geotropismus bei Thieren. *Arch. f. d. ges. Physiol.* Vol. xlix, pp. 175–189.

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